

## The Nutritional Status of Astronauts Is Altered after Long-Term Space Flight Aboard the International Space Station<sup>1</sup>

Scott M. Smith,<sup>\*2</sup> Sara R. Zwart,<sup>\*</sup> Gladys Block,<sup>†</sup> Barbara L. Rice,<sup>\*\*</sup> and Janis E. Davis-Street<sup>\*\*</sup>

<sup>\*</sup>Human Adaptation and Countermeasures Office, NASA Lyndon B. Johnson Space Center, Houston, TX 77058; <sup>†</sup>Epidemiology and Public Health Nutrition, University of California-Berkeley, Berkeley, CA 94720; and <sup>\*\*</sup>Enterprise Advisory Services, Inc., Houston, TX 77058

**ABSTRACT** Defining optimal nutrient requirements is critical for ensuring crew health during long-duration space exploration missions. Data pertaining to such nutrient requirements are extremely limited. The primary goal of this study was to better understand nutritional changes that occur during long-duration space flight. We examined body composition, bone metabolism, hematology, general blood chemistry, and blood levels of selected vitamins and minerals in 11 astronauts before and after long-duration (128–195 d) space flight aboard the International Space Station. Dietary intake and limited biochemical measures were assessed during flight. Crew members consumed a mean of 80% of their recommended energy intake, and on landing day their body weight was less ( $P = 0.051$ ) than before flight. Hematocrit, serum iron, ferritin saturation, and transferrin were decreased and serum ferritin was increased after flight ( $P < 0.05$ ). The finding that other acute-phase proteins were unchanged after flight suggests that the changes in iron metabolism are not likely to be solely a result of an inflammatory response. Urinary 8-hydroxy-2'-deoxyguanosine concentration was greater and RBC superoxide dismutase was less after flight ( $P < 0.05$ ), indicating increased oxidative damage. Despite vitamin D supplement use during flight, serum 25-hydroxycholecalciferol was decreased after flight ( $P < 0.01$ ). Bone resorption was increased after flight, as indicated by several markers. Bone formation, assessed by several markers, did not consistently rise 1 d after landing. These data provide evidence that bone loss, compromised vitamin D status, and oxidative damage are among critical nutritional concerns for long-duration space travelers. *J. Nutr.* 135: 437–443, 2005.

**KEY WORDS:** • space flight • nutritional status • humans • bone resorption • weightlessness

In a vigorous human space exploration program, with mission durations far exceeding any Space Shuttle or International Space Station (ISS)<sup>3</sup> mission duration to date, maintenance of crew member health will be of critical importance. Proper nutrition will be essential to this effort. In order to provide nutritional recommendations to crew members for long-duration space travel, we need to better understand how nutritional status and general physiology are affected by the microgravity environment. Dietary intake during space flight has often been inadequate (1–3), and this can greatly compromise nutritional status. Although some information is available about nutritional status during and after flight (1,4–7), the small sample sizes and incomplete data sets preclude a complete understanding of the role of nutrition in maintaining health in human space crews.

Data from both short- and long-duration space flights pro-

vide evidence that energy intake is typically 30–40% below the WHO recommendation, but energy expenditure is typically unchanged or even increased (1–3,8,9). This imbalance may explain some of the negative changes in overall nutritional status during flight. However, blood concentrations of some nutrients, such as vitamin D, continue to be low even when astronauts receive supplements during flight (4). Data from individual Skylab missions show that crew members on the longest mission (Skylab 4, 84 d), but not the shorter missions (28 and 59 d), had decreased serum 25-hydroxycholecalciferol [25(OH)-D<sub>3</sub>] at landing despite daily vitamin D supplementation (4). Similarly, in 2 separate studies, we reported that crew members on the Russian space station Mir had serum 25(OH)-D<sub>3</sub> concentrations that were 32–36% less during and after long-duration (3- to 4-mo) missions than before the missions (1,5,10). Ground-based studies of subjects living in closed-chamber facilities for extended periods also support these data (1).

The space environment itself results in physiologic changes that can alter nutritional status. For example, changes in iron metabolism are closely associated with hematological alterations during space flight (1,11,12). Similarly, increased levels of radiation and oxidative stress during flight likely contribute to decreased antioxidant status during or after space flight.

<sup>1</sup> Supported by NASA.

<sup>2</sup> To whom correspondence should be addressed.

E-mail: scott.m.smith@nasa.gov.

<sup>3</sup> Abbreviations used: 25(OH)D<sub>3</sub>, 25-hydroxycholecalciferol; 3-MH, 3-methyl-histidine; 8OHdG, 8-hydroxy-2'-deoxyguanosine; DEXA, dual-energy X-ray absorptiometry; GLA,  $\gamma$ -carboxyglutamic acid; GSH, glutathione; ISS, International Space Station; LBM, lean body mass; MCV, mean corpuscular volume; PTH, parathyroid hormone; SOD, superoxide dismutase; TAC, total antioxidant capacity.

In this study, we sought to better understand the nutritional status changes that occur during long-duration space flight. A secondary goal of this study was to determine whether changes in nutrient intake during flight were related to changes in nutritional status recorded after flight. The results presented here are data from astronauts who flew on 4- to 6-mo missions aboard the ISS. These data represent the first report of nutritional status from this space platform and the most complete nutritional assessment of space crews to date.

## SUBJECTS AND METHODS

**Subjects.** Subjects were U.S. astronauts on ISS Expeditions 1–8 (missions of 128 to 195 d during 2000–2004). The age of the 11 subjects (1 or 2 subjects per expedition, 2 females) was  $46.5 \pm 4.1$  y (mean  $\pm$  SD) before flight. For all but 2 of the crew members, preflight sample collections were conducted at the Johnson Space Center in Houston, Texas. The other 2 crew members' preflight sample collections were conducted in Star City, Russia; however, the time points from these collections were not different from the time points of the preflight sample collections conducted in the United States. Regardless of collection site, all samples were analyzed at the Johnson Space Center.

Following the loss of the Space Shuttle Columbia and subsequent grounding of the U.S. Space Shuttle program, crew members from 3 of the ISS Expeditions ( $n = 4$ ) landed in Russia instead of the United States. Thus, their postflight biological samples were collected in Star City, Russia. Postflight samples for the 5 expeditions ( $n = 7$ ) that landed in the United States were collected at the Kennedy Space Center in Florida. Regardless of collection site, all samples were analyzed at the Johnson Space Center. The protocol for this study was approved by the Johnson Space Center Committee for the Protection of Human Subjects.

**Food system.** The ISS food system provides a menu with a cycle of 6 to 10 d. About half of the food items are supplied by the United States and the other half are supplied by Russia (13). Foods are packaged in single-serving containers and are thermostabilized, dehydrated, irradiated, intermediate moisture, or natural form (13). Before each mission, crew members participate in food-tasting sessions, and dietitians plan menus that will use crew choices and best fulfill the defined nutritional requirements for space flight (14). These requirements have been derived from space flight research, extrapolated from speculation about the effects of space flight on nutrient needs, or applied directly from ground-based dietary reference intakes for micronutrients and WHO recommendations (15). A key concern for space flight, and limitation of the food system, is vitamin D. Accordingly, vitamin D supplements ( $10 \mu\text{g/d}$ ) were provided for the crew members. Some crew members consumed multivitamin supplements at their own discretion and/or in consultation with their flight surgeon.

**FFQ.** During flight, crew members were asked to record their dietary intake once per wk using an FFQ designed for use with the space flight food system. This FFQ has been validated in a ground-based model of long-duration space flight (1). Given the closed food system (with repetitive menu cycle), known portion sizes, and precise nutrient content for each food item in the system, the FFQ designed for space flight is much more reliable than a standard food questionnaire.

The FFQ is designed to obtain a near-real-time estimate of intakes of energy, protein, water, sodium, calcium, and iron, as well as to collect information about supplement use and any crew comments (16). The questionnaire input is transmitted to the ground, and results are calculated and reported to the flight surgeon within 24 to 48 h.

A unique FFQ was developed for each expedition to the International Space Station and was based on the specific menu for the crew on board and potential foods on board from earlier crews. Nutrient analyses by the NASA Johnson Space Center Water and Food Analytical Laboratory were used to categorize foods in the FFQ to optimize data from the nutrients of interest.

An additional ground-based validation of each FFQ was completed by comparing the nutrient analysis of the menu (using prox-

imate analysis data) with results obtained by entering the menu items into the FFQ. This was done at 2 levels, 100% (assumed complete menu consumption) or 66% (inadequate consumption).

**Body mass and body composition determinations.** Body mass was determined before, during, and after each flight; body composition (bone mineral content, bone mineral density, lean body mass, fat mass) was determined before and after each flight. Body mass and body composition before and after flight were determined by dual-energy X-ray absorptiometry (DEXA) with a fan beam densitometer (Hologic QDR 4500W, Hologic). Whole-body scans for body composition assessment were performed about 180 and 45 d before launch (designated L-180 and L-45) and 5 d after landing (designated Return + 5 d, or R + 5).

Body mass during flight was determined using a body mass-measuring device. The body mass-measuring device exerts a known force on the body, and body acceleration is measured. According to Newton's second law, body mass can be calculated from the force and acceleration. Body weight was also determined, using a standard clinical scale, before (L-180 and L-45) and after (landing day, R + 0) each flight.

**Biological sample collection and processing.** Preflight blood and initial urine samples were collected at about L-180 and L-45 for all crew members. For crew members landing in the United States, postflight samples were collected on R + 0 within 2 to 4 h of landing. For crew members on the expeditions that landed in Russia, postflight urine collection began on R + 1 or R + 2, and blood samples were collected 9 to 16 h after landing. Preflight blood samples were collected after an 8-h fast, but fasting did not always occur before collection of postflight blood samples. Crew members on the 5 Shuttle landings in the United States generally fasted 4 to 6 h before the R + 0 blood collection.

Blood samples were collected into appropriate tubes and processed to yield whole blood, plasma, or serum, depending on the specific analyte to be measured. A total of about 23.7 mL of blood was collected from each subject for all tests described herein.

During flight, blood samples were collected by finger stick for real-time analysis of blood pH and ionized calcium.

Pre- and postflight urine samples were collected over 48 h in individual bottles and stored in coolers until they were processed. Twenty-four-hour urine pools were created, pH was measured, and aliquots were prepared and frozen at  $-80^\circ\text{C}$  for subsequent analysis.

**Biochemical analyses.** Regardless of sample collection site, analyses were performed at the Johnson Space Center by trained personnel. Most analyses were performed by standard commercial techniques, and all have been previously described in detail (1,17).

**Statistical analysis.** Statistical analyses were designed to test the hypothesis that nutritional status was different postflight compared to preflight. We accounted for the difference in landing site in a subset of crew members, because the timing of sample collections in those crew members is a potential confounding factor. We also controlled for duplicate preflight sessions in crew members. Details of the approach used are described herein. Because all sample analyses were performed at the Johnson Space Center, any effect of landing site is not related to sample analysis, but likely to the time of sample collection (i.e., number of hours from touchdown) that varied between the 2 sites.

Statistical analyses were performed with the data in their original form or on a transformed (reciprocal, square, or natural logarithm) scale to achieve normality and homogeneity of variability as determined by the Kolmogorov-Smirnov normality test. Data for some variables [RBC folate, body mass, 8-hydroxy-2'-deoxyguanosine (8OHdG), and serum selenium] could not be normalized; in these cases, the nonnormalized data were analyzed.

Student's *t* test was used to analyze for differences between the 2 preflight collection times (L-180 and L-45). If no differences were noted (as occurred in all but 1 case), preflight mean values were determined and compared with postflight (R + 0 through R + 2) data using a two-way repeated measures ANOVA, with time and landing site (United States and Russia) as repeated factors. The dependent variables were the analytes measured. Post hoc Bonferroni tests were performed to assess specific differences between times or landing sites. For the case where L-180 and L-45 values were signif-

icantly different, L-45 (instead of the preflight mean) was compared with the postflight data using two-way repeated measures ANOVA. These cases are noted in the Results.

In instances where significant outliers existed (as determined by Grubbs' test), the data were analyzed with and without the outlier and both results were reported. For some analyses in which correlations were assessed, the data were analyzed by simple linear regression and a Pearson correlation coefficient ( $r$ ) was calculated. Statistical analyses were performed using SigmaStat software 3.01a (SPSS), and  $P < 0.05$  was the level of significance. Data are expressed as means  $\pm$  SD.

## RESULTS

**FFQ.** Completed FFQs were received  $46 \pm 28\%$  of the weeks on orbit (range 6–95%). There are many possible reasons why the FFQ was not completed for any given week, but schedule and time constraints were primary causes. Additionally, during 2 of the early expeditions, a software error reduced the number of completed FFQs received.

One way to validate the FFQ was to calculate the results obtained from entering the planned menu contents for each Expedition into the FFQ (at 100 or 66% of menu content, for high and low reference points) and compare the FFQ result to the exact nutrient data from proximate analysis of the same menu foods. The FFQ estimated the intake of energy within  $97 \pm 5\%$  of proximate analysis at 100% intake and  $97 \pm 7\%$  at 66% intake. Similar results were found for other nutrients (data not shown).

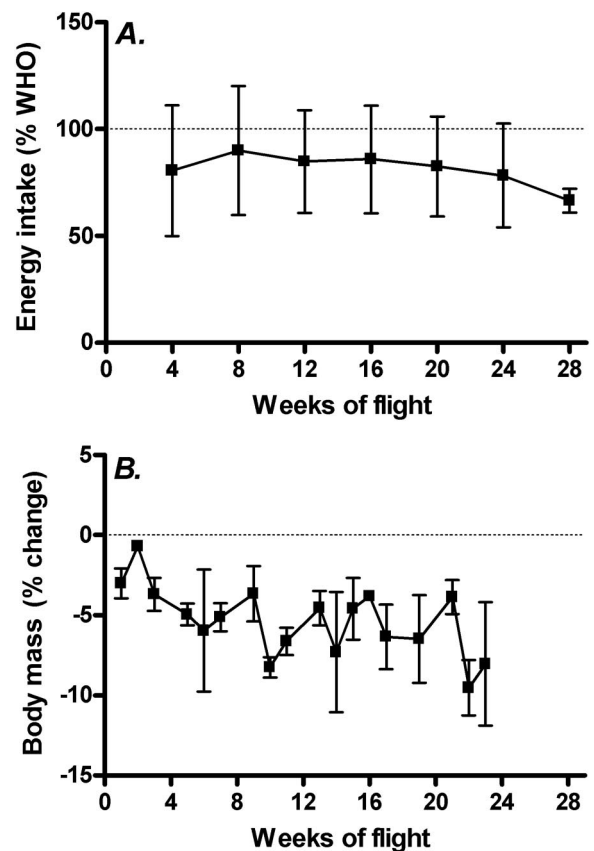
**Food intake.** The mean energy intake based on the FFQ for the entire in-flight period ( $n = 11$ ) was  $2284 \pm 627$  kcal ( $9563 \pm 2625$  kJ), which is equivalent to  $80 \pm 21\%$  of the WHO recommendation (Fig. 1A). Total protein intake during flight was  $102 \pm 29$  g, sodium intake was  $4556 \pm 1492$  mg, calcium intake was  $1068 \pm 384$  mg, and iron intake was  $23 \pm 12$  mg. During flight, subjects reported consuming  $5.7 \pm 4.0$  vitamin D supplements per week (each supplement contained  $10 \mu\text{g}$  cholecalciferol, and this number accounts for the vitamin D from any multivitamin consumed). Subjects consumed a mean of  $3.5 \pm 2.9$  multivitamin supplements per week.

In several situations during missions, concerns were raised about inadequate intake of nutrients (most often energy). Recommendations were made to the flight surgeon (i.e., the physician assigned to each crew) regarding potential means of increasing intake, including highlighting food items that were more energy dense and items that the crew member had previously reported consuming (to avoid recommending foods that were not liked). A crew member who received dietary counseling was able to consume the recommended energy intake during flight (Fig. 2).

**Body composition.** Body weight had decreased about 5% ( $P = 0.051$ ) on landing day (R + 0) (Table 1). In-flight body mass results (expressed as a percentage change from preflight values) are shown in Figure 1B. Because of the small number of subjects, differences in data collection schedules, and differences between instruments used to measure body mass during flight and on the ground, statistical analyses were not performed on the in-flight data.

Both total bone mineral content and bone mineral density were lower on landing day than before flight ( $P < 0.01$ ) (Table 1). Neither lean body mass nor fat mass was different after flight.

**Oxidative stress.** The urinary concentration of 8OHdG was elevated about 32% after landing ( $P < 0.05$ ), indicating that increased DNA damage was present after space flight (Table 2). RBC superoxide dismutase was less after landing, indicating a decreased antioxidant capacity during flight. Al-

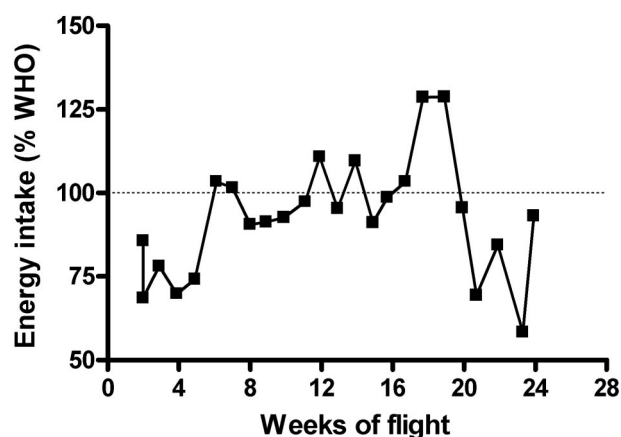


**FIGURE 1** In-flight energy intake and body mass. A. Mean energy intake during space flight expressed as a percentage of the WHO recommendation (15). On average, energy intake data were available for each subject every 1 to 2 wk. Data are means  $\pm$  SD for all available energy intake data over successive 4-wk intervals ( $n = 15, 17, 18, 16$ , and 11 for each 4-wk interval, in chronological order). B. In-flight body mass expressed as a percentage change from preflight mass. Data are weekly means  $\pm$  SD. Mass data were not available for all 11 ISS crew members each week; the data presented are all of the data available for each week.

though postflight total antioxidant capacity (TAC) was not different from preflight TAC, on landing day 6 of the 11 crew members had TAC values below the low end of the normal clinical range ( $1.285 \text{ mmol/L}$ ) (individual data not shown). Malondialdehyde concentration was not changed after landing.

**General chemistry, vitamin, and mineral measurements.** Routine clinical chemistry variables were generally unchanged after landing compared to before launch (Table 2). Preflight and postflight urinary 3-methylhistidine (3-MH), creatinine, pH, serum cholesterol, triglycerides, and blood pH did not differ. Three of the 7 crew members who landed in the United States had urinary iodine concentrations above the normal clinical range ( $3.6 \mu\text{mol/d}$ ) (individual data not shown). This was likely due to the consumption of iodinated water on the Space Shuttle in the final days before returning to Earth (as opposed to the Russian Soyuz vehicle, which does not provide iodinated water). Independent of landing site, serum selenium was lower after landing than before launch ( $P < 0.01$ ). Similarly, urinary magnesium and phosphorus were 44 and 46% lower after landing than before launch ( $P < 0.001$ ). Fifty-five percent of crew members had postflight urinary magnesium concentrations lower than the low end of the clinical range ( $3.0 \text{ mmol/d}$ , individual data not shown). The serum concen-





**FIGURE 2** Energy intake of 1 subject during flight, expressed as a percentage of the WHO requirement. Dietary counseling was provided for this subject after wk 5 because energy intake had been consistently low. At landing, this subject's weight was not substantially less (<2 kg) than it was before flight (<3% body weight).

tration of zinc tended to be lower ( $P = 0.06$ ) after flight than before flight.

Serum concentrations of retinyl palmitate were significantly greater after landing than before launch (Table 2). Serum  $\gamma$ -tocopherol concentration was 46% less after long-duration space flight than before flight ( $P < 0.05$ ), but  $\alpha$ -tocopherol was unchanged. Phylloquinone was 42% less after flight than before flight ( $P < 0.01$  for normalized data). Of the water-soluble vitamins assessed, RBC folate concentrations were about 20% less ( $P < 0.01$ ) after landing. Qualitative RBC transketolase data (with one exception preflight) were within the normal range (<15% activation) (18).

**Bone markers.** The vitamin D status indicator 25(OH)-D<sub>3</sub> was 25% less after landing than before flight ( $P < 0.01$ ), with concentrations ranging from 17 to 92 nmol/L (Table 3). The concentration of 1,25-dihydroxycholecalciferol, the active form of vitamin D, was not different after landing, although the serum concentration at landing for crew members with Russian landings tended to be greater than that

**TABLE 1**

*Body composition of astronauts before and after long-duration space flight<sup>1,2</sup>*

	Preflight	R + 0/5
Bone mineral content, kg	2.81 $\pm$ 0.43	2.73 $\pm$ 0.42***
Bone mineral density, g/cm <sup>3</sup>	1.27 $\pm$ 0.11	1.24 $\pm$ 0.12**
Body weight, kg		
DEXA	74.3 $\pm$ 6.1	72.5 $\pm$ 7.4
Calibrated scale	75.4 $\pm$ 6.2	72.7 $\pm$ 8.0#
LBM, kg	56.2 $\pm$ 7.2	55.1 $\pm$ 8.3
LBM, %	75.4 $\pm$ 5.2	75.7 $\pm$ 5.7
Fat mass, kg	15.3 $\pm$ 3.6	14.7 $\pm$ 3.4
Fat, %	20.8 $\pm$ 5.5	20.5 $\pm$ 5.9

<sup>1</sup> Data are means  $\pm$  SD,  $n = 11$ . Preflight data are means of L-180 (launch minus 180 d) and L-45. Symbols indicate a significant effect of time, \*  $P < 0.05$ , \*\*  $P < 0.01$ , \*\*\*  $P < 0.001$ ; #  $P = 0.051$  when the preflight body weight mean was used and  $P = 0.050$  when only L-45 body weight was compared with R + 0.

<sup>2</sup> Body weights on a calibrated scale were determined on R + 0; postflight values for all other variables were determined on R + 5.

**TABLE 2**

*General chemistry, vitamins, minerals, and antioxidant/oxidative damage markers of astronauts before and after long-duration space flight<sup>1</sup>*

	Preflight	R + 0
Urine		
3-MH, <sup>2</sup> $\mu$ mol/d	289.7 $\pm$ 60.6	247.9 $\pm$ 127.3
Creatinine, mmol/d	15.2 $\pm$ 2.1	15.2 $\pm$ 3.0
Iodine, <sup>3</sup> $\mu$ mol/d	3.08 $\pm$ 2.04	3.29 $\pm$ 2.10††
Magnesium, mmol/d	4.8 $\pm$ 1.8	2.7 $\pm$ 0.8***
Phosphorus, mmol/d	31.5 $\pm$ 8.4	16.9 $\pm$ 5.6***
8OHdG, nmol/mmol creatinine	82.5 $\pm$ 24.1	107.8 $\pm$ 28.1*
GLA, <sup>4</sup> $\mu$ mol/mmol creatinine	2.32 $\pm$ 0.54	2.62 $\pm$ 1.50
pH	6.02 $\pm$ 0.28	6.06 $\pm$ 0.51
Serum		
pH (blood)	7.37 $\pm$ 0.02	7.37 $\pm$ 0.05
Copper, $\mu$ mol/L	16.09 $\pm$ 2.67	15.31 $\pm$ 5.12
Zinc, $\mu$ mol/L	20.4 $\pm$ 3.9†	17.02 $\pm$ 3.44
Selenium, $\mu$ mol/L	2.29 $\pm$ 0.27	2.03 $\pm$ 0.22**†
Cholesterol, mmol/L	4.80 $\pm$ 0.65	4.81 $\pm$ 1.05
Triglycerides, mmol/L	0.80 $\pm$ 0.28	0.75 $\pm$ 0.24
Albumin, g/L	44 $\pm$ 1.0	44 $\pm$ 4.0†
Glutathione peroxidase, U/g		
hemoglobin	48.7 $\pm$ 12.0	49.3 $\pm$ 10.3
Malondialdehyde, $\mu$ mol/L	1.07 $\pm$ 0.45	0.68 $\pm$ 0.50
TAC, mmol/L	1.43 $\pm$ 0.14	1.30 $\pm$ 0.19
SOD, U/g hemoglobin	1315 $\pm$ 101	1195 $\pm$ 132*
GSH reductase, % activation	18.2 $\pm$ 11.2	19.4 $\pm$ 15.3
Ceruloplasmin, mg/L	345 $\pm$ 63	355 $\pm$ 123
Retinol binding protein, mg/L	53.0 $\pm$ 10.4	50.7 $\pm$ 9.3†
Transferrin, mg/L	290 $\pm$ 47	300 $\pm$ 36
RBC transaminase, % activation	96.3 $\pm$ 19.8	97.9 $\pm$ 20.5
$\beta$ -Carotene, $\mu$ mol/L	0.40 $\pm$ 0.42	0.45 $\pm$ 0.28
RBC folate, <sup>5</sup> nmol/L	1549 $\pm$ 403	1260 $\pm$ 423**†
Retinol, $\mu$ mol/L	2.09 $\pm$ 0.57	2.07 $\pm$ 0.47†
Retinyl palmitate, nmol/L	30.1 $\pm$ 12.8	61.5 $\pm$ 33.6**†
$\gamma$ -Tocopherol, $\mu$ mol/L	3.2 $\pm$ 1.9	1.6 $\pm$ 1.1*
$\alpha$ -Tocopherol, $\mu$ mol/L	30.1 $\pm$ 8.4	32.5 $\pm$ 6.7
Phylloquinone, nmol/L	1.2 $\pm$ 0.6	0.7 $\pm$ 0.5*

<sup>1</sup> Data are means  $\pm$  SD,  $n = 11$ . Preflight data are means of L-180 (launch minus 180 d) and L-45. Symbols indicate a significant effect of time, \*  $P < 0.05$ , \*\*  $P < 0.01$ , \*\*\*  $P < 0.001$ ; symbols indicate a significant interaction between time and landing site, †  $P < 0.05$ , ††  $P < 0.01$ ; and ‡ L-180 was different from L-45 (for these cases, the L-45 value is reported rather than the preflight mean).

<sup>2</sup> An outlier was identified for the R + 0 value, but excluding the outlier yielded a significant effect of time. The results presented include all of the data.

<sup>3</sup> An outlier was identified for the preflight mean. Excluding the outlier from the statistical analysis yielded a significant interaction term but no effect of landing site. The results presented include all of the data.

<sup>4</sup> An outlier was identified for the R + 0 value, but excluding this value did not alter the statistical results.

<sup>5</sup> An outlier was identified for the preflight mean; excluding this outlier did not change the main effect, but there was no longer a significant interaction.

of crew members with U.S. landings ( $P = 0.053$ ). Unlike previous space flight findings, urinary calcium at landing did not differ ( $P = 0.50$ ) from that before launch, but the blood concentration of ionized calcium was lower ( $P = 0.06$ ) after landing than before launch. Eight of the 11 crew members had blood ionized calcium concentrations at or below the lower limit of the normal clinical range (1.19 mmol/L) (individual data not shown). All markers of bone resorption that were measured were significantly greater after landing than before launch (Table 3). The excretion of deoxypyridinoline was 75% greater ( $P < 0.01$ ), excretion of N-telopeptide was about

TABLE 3

*Serum and urine calcium and bone markers of astronauts before and after long-duration space flight<sup>1</sup>*

	Preflight	R + 0
25(OH)-cholecalciferol, nmol/L	63.4 ± 15.8	47.7 ± 20.4**
1,25(OH) <sub>2</sub> -cholecalciferol, pmol/L	124.5 ± 37.3	121.1 ± 38.9†
Serum calcium, mmol/L	2.34 ± 0.03	2.30 ± 0.10
Serum ionized calcium, mmol/L	1.21 ± 0.02	1.17 ± 0.05
Urinary calcium, mmol/d	4.44 ± 1.79	4.10 ± 1.79
Bone-specific alkaline phosphatase, U/L	17.6 ± 2.9	24.9 ± 5.5***†
Osteocalcin, µg/L	11.2 ± 2.7	12.2 ± 3.4†
Alkaline phosphatase, U/L	52.5 ± 11.0	60.9 ± 10.5*
PTH, ng/L	29.2 ± 8.0	32.7 ± 12.4
Urinary deoxypyridinoline, nmol/d	45.2 ± 19.1	79.1 ± 19.5**
Urinary N-telopeptide, nmol/d	408 ± 109	567 ± 179***
Urinary pyridinium crosslinks, nmol/d	230 ± 46	365 ± 150*

<sup>1</sup> Data are means ± SD, *n* = 11. Preflight data are means of L-180 (launch minus 180 d) and L-45. All measurements are serum levels unless otherwise indicated. Symbols indicate a significant effect of time \* *P* < 0.05, \*\* *P* < 0.01, \*\*\* *P* < 0.001; † significant interaction between time and landing site, *P* < 0.05.

<sup>2</sup> An outlier was identified for R + 0, but the statistics were unchanged when this outlier was removed. The results presented include all of the data.

<sup>3</sup> An outlier was identified for the preflight mean, but the statistics were unchanged when this outlier was removed. The results presented include all of the data.

40% greater (*P* < 0.001), and excretion of pyridinium crosslinks was 60% greater (*P* < 0.05) after landing than before launch. The timing of sample collection after flight may affect indicators of bone formation, because bone-specific alkaline phosphatase was greater for astronauts with Russian landings than for those with U.S. landings (*P* < 0.05) (Table 3). For osteocalcin, values from Russian landings were less after flight than before flight, and values from U.S. landings were greater after flight than before flight. Before launch, 25(OH)-D<sub>3</sub> was inversely correlated with parathyroid hormone (PTH) (*r* = -0.72, *P* < 0.05), but this relation was not evident after landing.

**Hematologic variables.** The observed hematologic changes reflected the expected response to space flight, with reduced hemoglobin concentration (*P* < 0.05) and hematocrit (*P* < 0.01) and increased serum ferritin (*P* < 0.01) (Table 4). Ferritin iron saturation (*P* < 0.05) and mean corpuscular volume (MCV) (*P* < 0.05) were lower after landing (Table 4). There was no relation between iron intake and any of the hematologic changes.

## DISCUSSION

The data presented herein provide evidence of significant changes in nutritional status after space flight, some likely related to inadequate dietary intake and others clearly related to effects of space flight. Low energy intake remains a critical issue for crew members because it is generally associated with inadequate vitamin and mineral intake. Similar to previous studies (1–3), energy intake was significantly below the WHO recommendation in this study. Based on previous metabolic studies from Skylab, it is possible for crew members to consume 100% of their recommended energy intake (4). The low energy intake may be related to time required for meal preparation or time allotted for meals.

Vitamin D status was altered after long-duration space

flight (about 4 to 6 mo). Despite the reported use of vitamin D supplements by some of the astronauts (5.7 ± 4.0 supplements/wk), the serum concentration of 25(OH)-D<sub>3</sub> for the ISS crew members in this study was about 25% less after landing than before launch. Reported supplement use was not related to 25(OH)-D<sub>3</sub> status (regression data not shown).

The normal range for serum 25(OH)-D<sub>3</sub> is about 23 to 117 nmol/L, but the lower limit can vary from 20 to 37.5 nmol/L depending on the geographic location where the blood samples were collected (19). The cutoff point to define a vitamin D deficiency is a source of some controversy because past recommendations have been based solely on preventing rickets or osteomalacia and not on chronic deficiency diseases (20). It is now evident that vitamin D status is an important determinant for long-latency diseases as well and that marginal deficiencies should be considered as well as frank deficiencies (21–24). Several studies suggest that an optimal serum 25(OH)-D<sub>3</sub> level is around 80 nmol/L (23,25,26), which would mean that 80–90% of the subjects had suboptimal vitamin D status before and after their flights. Not only was vitamin D status decreased after long-duration space flight, but also vitamin D metabolism or function may have been altered. Serum 25(OH)-D<sub>3</sub> concentration is inversely proportional to serum PTH (20,27), and this relation was evident among crew members before flight but not after flight. The difference between the preflight and postflight relation of PTH and 25(OH)-D<sub>3</sub> suggests that the body's normal response to changes in vitamin D status was altered.

Altered vitamin D status after landing was accompanied by evidence of increased bone resorption. After landing, urinary markers of bone resorption were 40–75% greater than before launch; this supports previous findings (5,7,10,28,29).

No previous evidence exists for increased bone formation during flight, but increases have been noted after landing (5,10). Likewise, bone formation is unchanged during bed rest (a ground-based model of weightlessness) (30–33), but evidence suggests that bone formation increases after reambulation (34,35). The differences in timing of the sample collections at the Russian and U.S. landing sites may explain the significant effect of landing site on bone marker data.

Urinary magnesium and phosphorus concentrations were about 45% less after landing than before launch. Results of previous space flight studies are consistent with a significant decrease in urinary magnesium (4,36), possibly owing to a

TABLE 4

*Hematologic variables and iron status of astronauts before and after long-duration space flight<sup>1</sup>*

	Preflight mean	R + 0
Hematocrit	0.42 ± .03	0.40 ± .03**
Hemoglobin, g/L	140 ± 10	135 ± 10*
MCV, fL	90.9 ± 3.1	89.3 ± 2.8*
Ferritin, µg/L	94.8 ± 71.7	138.8 ± 66.0**
Ferritin iron <sup>2</sup>		
µmol/L	0.26 ± 0.07	0.32 ± 0.14
% saturation	20.3 ± 9.0	13.8 ± 4.4*
Serum iron, µmol/L	16.8 ± 3.3	11.1 ± 2.9**
Transferrin, g/L	2.54 ± 0.25	2.27 ± 0.19**
Transferrin receptors, mg/L	4.3 ± 0.9	3.9 ± 0.8

<sup>1</sup> Data are means ± SD, *n* = 11. Preflight data are means of L-180 (launch minus 180 d) and L-45. Symbols indicate a significant effect of time, \* *P* < 0.05, \*\* *P* < 0.01.

<sup>2</sup> An outlier was identified for the preflight mean, but excluding this value did not alter results. The results presented include all of the data.

decrease in magnesium intake. Decreased urinary magnesium could be a point of concern for long-duration flights because of the role of magnesium in inhibiting calcium oxalate renal stones (37,38).

The role of vitamin K in space flight must be further evaluated because this study and others suggest that the serum concentration of vitamin K is less during space flight than before flight, and a decreased vitamin K status has negative implications for bone health (39). Vitamin K is involved in the formation of  $\gamma$ -carboxyglutamic acid (GLA) in proteins, such as osteocalcin and matrix GLA protein. Although the urinary excretion of GLA did not change significantly in this study, serum phyloquinone was 42% less ( $P < 0.05$ ) after landing than before launch. Other studies demonstrate that vitamin K supplementation during space flight significantly elevates urinary GLA excretion and decreases the excretion rate of undercarboxylated osteocalcin, suggesting that vitamin K status is compromised during space flight (40,41).

Folate and vitamin E are also affected by space flight. The results of the present study provide no evidence that vitamin B-6, riboflavin,  $\beta$ -carotene, or retinol was altered by exposure to microgravity. RBC folate decreased about 20% ( $P < 0.01$ ). Before launch, the RBC folate of most subjects was at or near the upper limit of the normal range. After landing, the RBC folate of many subjects approached the lower limit of the normal range, and it is not known whether this decrease in folate status would level off or continue to decrease with missions of even longer duration. Food processing techniques do not reduce the availability of folate in the space flight diet (42). Thus inadequate food intake rather than limited folate availability during flight is likely a key factor contributing to the decrease in folate status. The number of multivitamins consumed per week was positively correlated ( $r = 0.62$ ,  $P < 0.05$ ) with RBC folate levels (data not shown), which suggests that the decreased folate status during flight is likely related to decreased folate intake.  $\gamma$ -Tocopherol but not  $\alpha$ -tocopherol was significantly decreased after landing. This may have been caused by decreased overall food intake or radiation/oxidative stress. The unchanged  $\alpha$ -tocopherol may relate to supplement use during flight.

Several oxidative stressors in the space flight environment have the potential to induce oxidative damage to biological systems. Two such stressors are radiation hazards and periods of exposure to hyperoxia. The decreased superoxide dismutase (SOD) after landing suggests that antioxidant capacity decreases during flight. Although there was no evidence that lipid peroxidation occurred, serum 8OHdG was greater after landing than before launch, suggesting that the amount of DNA damage was greater after flight. The magnitude of the increase in 8OHdG is similar to the effects of both active cigarette smoking and passive exposure to environmental cigarette smoke (43,44).

The findings reported here for hematologic variables and iron indices support previous preliminary data obtained from 2 subjects before and after long-duration space flight (1). The significant postflight decreases in hemoglobin, hematocrit, and MCV are consistent with previously reported RBC mass reduction (11,12). Along with findings after landing of significantly increased amounts of ferritin and decreased amounts of transferrin and slightly greater amounts of ferritin iron, these data suggest that iron metabolism is altered and that storage pools of iron may be shifted as a result of exposure to microgravity (45). Because ferritin concentration increased and serum iron concentration decreased, the possibility that these changes were caused by an acute inflammatory response cannot be ruled out (46). However, the finding that other acute-

phase proteins (such as ceruloplasmin) did not change suggests that several mechanisms may be involved.

The data reported here provide evidence that nutritional status is altered during and after long-duration space flight. Four decades of human space flight have shown that many physiological systems are affected by space travel, and this has many implications for nutrition. Nutrients can be casualties of these effects, and in some cases they may be used as countermeasures to these effects. Moreover, additional efforts are required to better understand the role of nutrition in bone health and changes in body composition. Maintaining and monitoring nutritional status are important for ensuring crew health during space flight and will be critical as we begin to embark on longer duration exploration missions in the future.

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